

SHORT COMMUNICATION

Influence of morphological, chemical and physical leaf traits on food selection of a herbivorous iguana from The Bahamas

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Abstract: Herbivores are predicted to forage on a variety of plants in order to obtain a nutritionally sufficient diet. Most herbivores, however, forage non-randomly and may be influenced by morphological, chemical and physical traits in their food. We examined the influence of several leaf traits on food selection for the Exuma rock iguana (*Cyclura cyclura figginsi*). We expected the iguana to prefer leaves with higher nutrient concentration and lower physical defences, such as reflected by high N, P, Ca, K, Mg concentrations and low leaf density and per cent concentrations of cellulose, hemicellulose and lignin, respectively. We quantified selection by examining 30 faecal samples and analysing traits of leaves from the 10 most common plants on the island. Our results showed substantial variability in all measured traits among species but food preference only for less-dense leaves, a good indicator of low leaf toughness. Our results are the first to demonstrate that physical leaf traits can influence food selection in a true herbivorous lizard and offer a basis for future testing.

Key Words: *Cyclura cyclura*, herbivory, Iguanidae, island, leaf density, lizard

To obtain a nutritionally adequate diet, generalist herbivores are predicted to forage on a variety of plant species (Westoby 1974). Most free-ranging herbivores, however, forage non-randomly in the presence of a broad assortment of plants (Marsh *et al.* 2006). Thus, while nutrient concentrations in leaves may drive food selection in some herbivorous species (Pérez-Harguindeguy *et al.* 2003), other factors such as toxic secondary compounds (Freeland & Janzen 1974), foliar water content (Poorter *et al.* 2004) or leaf mechanical strength (Kitajima & Poorter 2010) can also influence selection. In herbivorous lizards, little is known about what prompts their foraging decisions, although it has been shown that they can exhibit specific food preferences under natural conditions (Dearing & Schall 1992, van Marken Lichtenbelt 1993). Understanding food-selection behaviour in herbivorous lizards, a specialized group comprising approximately 1% of lizard species (Cooper & Vitt 2002), is important given their role as structural modifiers of vegetation

communities (Auffenburg 1982) and as potentially significant seed dispersers (Moura *et al.* 2015).

In this study we examined the influence of leaf traits on food selection of a free-ranging herbivorous lizard, the Exuma rock iguana (*Cyclura cyclura figginsi* Barbour 1923), by quantifying selection using faecal examinations, and relating it to leaf traits (leaf area and density), chemical concentrations of nutrients and fibre fractions. Since it is expected that generalist herbivores require a variety of elements to fulfil their nutritional demands, we hypothesized that the iguana would not select plants based on any singular foliar nutrient concentration but would prefer leaves with overall higher nutrient concentration. Additionally, high material strength, such as those conferred by fibrous carbon polymers, can decrease food digestibility and minimize foraging efficiency (Clissold *et al.* 2009). For example, hemicellulose provides pliable cross-links between cellulose microfibrils, whereas cellulose influences the directional tensile strength of primary and secondary cell walls, and lignin provides permanent and rigid cross-linkages within the secondary cell wall (Kitajima *et al.* 2012). Thus, we expected iguanas to

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select leaves with low tissue density and low per cent fibre fractions.

The Exuma rock iguana is a large-bodied (max. snout–vent length and body mass: 58.0 cm and 7.94 kg, respectively) herbivorous lizard inhabiting cays in the Exuma Islands chain, Bahamas. We conducted research on Gaulin Cay (24°07′18.32″N; 76°24′19.57″W), a 13.6-ha island with a vegetation community consisting of 30 species. In May 2010, we captured iguanas using fish-landing nets and by noose. All captured iguanas were measured (snout–vent length, body mass) and sexed by cloacal probing for hemipenes. Faecal samples were collected from 30 iguanas primarily during processing. If iguanas did not defecate during processing, they were kept overnight in opaque cloth bags and samples collected the next morning. Faecal samples were examined and visible plant remains identified to species using Correll & Correll (1982).

We estimated plant species abundance using a 1-m² quadrat placed every 10 m along nine 100-m-long transects. We estimated preferences for each food type by calculating electivity values defined by Ivlev (1961): $\text{Electivity} = [r(i) - p(i)]/[r(i) + p(i)]$, where $r(i)$ is the proportion of food type i in the diet and $p(i)$ is the proportion of food type i in the environment. The result is a metric ranging from -1 (food considered strongly avoided) to 0 (food taken in its proportion in the environment) to $+1$ (food considered strongly preferred).

We focused our dietary analysis on leaves because seasonal fruits are reported eaten regardless of plant species whereas leaves are not (Auffenberg 1982). Leaves also represent a consistent, available food supply during our restricted sample period and remain intact through the digestive system. To study physical and chemical properties of foliage, we selected the 10 most common plant species and collected leaf samples from five plants per species. We calculated mean leaf area from 10 scanned leaves per species using Scion Image software (Scion Corporation, USA). We measured leaf thickness from the same fresh samples using a micrometer. Leaf volume was calculated as mean leaf area \times mean leaf thickness. Samples were then dried at 60°C and mean leaf density calculated as leaf dry weight/leaf volume. We used leaf density as a proxy for leaf material toughness (resistance to fracture per unit fracture area) because they are positively correlated ($r = 0.54$) in 197 species of tree and shrub species (Westbrook *et al.* 2011) and because leaf density is a good indicator of physical defence (Kitajima & Poorter 2010).

We measured nutrient and fibre concentrations using additional foliage from the collected samples. We measured total per cent N, with an elemental analyser (Costech Analytical, USA), and per cent P using ash digestion (Jones & Case 1996) followed by colorimetric determination of ortho-phosphate

using a spectrophotometer microplate reader (Bio-Tek Instruments Inc., USA). We measured per cent Ca, K, Mg using ICP spectroscopy. As an indicator of digestibility, we measured fibre fractions on leaves (per cent hemicellulose, cellulose and lignin), using a series of increasingly aggressive extractants in a fibre analyser system (ANKOM Technology, USA; Alvarez-Clare & Kitajima 2007).

To test for differences in physical and chemical properties among plant species, we conducted Kruskal–Wallis non-parametric tests. We evaluated the relationship among species means using pairwise Spearman correlations. We conducted linear regressions to test if mean foliar physical and chemical traits were good predictors of selection. We included biologically important ratios (Mans & Braun 2014) in our analyses but do not report results as none were statistically significant.

We identified one to three plant species in each faecal sample (mean \pm SD = 1.6 ± 0.7 species) and did not detect a significant relationship between iguana size and number of items in faecal samples ($r^2 = 0.04$, $P = 0.40$). All measured morphological and chemical leaf traits varied significantly among plant species but only N and P concentrations (Spearman $\rho = 0.78$, $P = 0.01$), and N and K concentrations (Spearman $\rho = 0.82$, $P < 0.01$) were correlated across species (Table 1). In addition, from all measured leaf traits only leaf density was a predictor of selection as iguanas preferred leaves with lower tissue density ($r^2 = 0.38$, $P = 0.05$).

Our results demonstrate substantial variability in nutritional traits among potential food types for iguanas, but no apparent selection toward any specific nutritional trait in leaves. As predicted, our data suggest that food selection over the short term is not driven ultimately by a singular nutritional component. Similar findings have been reported for other herbivorous lizards regarding nutritional (Blázquez & Rodríguez-Estrella 2007) and caloric (Auffenberg 1982) values. Together, these results support the nutritional constraint model, which purports that no one plant species can satisfy nutritional demands of a herbivore generalist (Westoby 1978) because plant tissues are relatively low in essential nutrients and digestible energy compared with those of animals (Robbins 1993).

Our results provide evidence that leaf material toughness (i.e. leaf density) can influence food selection in herbivorous iguanas over the short term and are consistent with studies conducted on invertebrates (Coley 1988) and primates (Teaford *et al.* 2006). Although we did not find a direct correlation between selection and the individual fibre fractions (% hemicellulose, % cellulose and % lignin), this is not surprising given that leaf toughness is not conferred exclusively by the chemical composition of the leaf but by the structural combination of these heterogeneous materials (Kitajima *et al.* 2012). Leaf density has been shown to be a good predictor of

Table 1. Mean (\pm SE) measured leaf traits for Bahamian plants listed by increasing preference (e.g. electivity index (EI)) by the Exuma rock iguana. Included are non-parametric Kruskal–Wallis H statistics comparing leaf traits across species. Plant species follow nomenclature of Correll & Correll (1982). Hemi, hemicellulose; Cellul, cellulose; * 0.05 > P > 0.001, ** P < 0.001.

Species	EI	Leaf area (cm ²)	Leaf density (g cm ⁻³)	% Hemi	% Cellul	% Lignin	N (mg g ⁻¹)	P (mg g ⁻¹)	K (mg g ⁻¹)	Ca (mg g ⁻¹)	Mg (mg g ⁻¹)
<i>Jacquinia keyensis</i>	-1.00	25.3 \pm 2.25	0.44 \pm 0.03	12.2 \pm 1.15	11.0 \pm 0.98	16.9 \pm 0.78	6.98 \pm 0.23	0.51 \pm 0.09	7.05 \pm 1.01	14.7 \pm 3.19	3.32 \pm 0.26
<i>Casasia clusifolia</i>	-1.00	209 \pm 23.7	0.33 \pm 0.04	8.59 \pm 1.06	6.37 \pm 1.64	13.3 \pm 1.28	7.49 \pm 0.76	0.57 \pm 0.10	8.79 \pm 0.86	12.7 \pm 0.31	5.26 \pm 0.76
<i>Manilkara bahamensis</i>	-1.00	14.2 \pm 22.7	0.36 \pm 0.03	8.15 \pm 1.13	17.2 \pm 0.28	17.0 \pm 1.31	12.2 \pm 0.88	0.82 \pm 0.09	9.32 \pm 0.43	8.05 \pm 1.30	3.04 \pm 0.27
<i>Rhachicallis americana</i>	-0.62	0.67 \pm 0.09	0.34 ^a	5.64 \pm 0.23	9.55 \pm 0.41	17.8 \pm 0.48	5.82 \pm 0.33	0.21 \pm 0.01	5.68 \pm 0.49	14.2 \pm 0.79	4.03 \pm 0.22
<i>Erithalis fruticosa</i>	-0.03	63.7 \pm 6.48	0.40 \pm 0.02	5.99 \pm 0.65	4.45 \pm 0.42	11.6 \pm 0.45	7.41 \pm 0.43	0.36 \pm 0.04	8.09 \pm 1.08	13.5 \pm 1.27	3.88 \pm 0.44
<i>Suriana maritima</i>	0.00	7.48 \pm 0.32	0.24 \pm 0.02	13.2 \pm 0.97	5.66 \pm 0.04	12.4 \pm 0.30	8.80 \pm 0.92	0.99 \pm 0.16	5.98 \pm 0.36	22.7 \pm 3.74	4.84 \pm 0.55
<i>Strunpfitia maritima</i>	0.00	3.60 \pm 0.29	0.27 \pm 0.01	3.88 \pm 0.44	6.28 \pm 0.54	22.4 \pm 0.62	4.64 \pm 0.14	0.29 \pm 0.06	5.76 \pm 0.39	13.3 \pm 0.75	2.45 \pm 0.39
<i>Conocarpus erectus</i>	0.46	98.0 \pm 11.1	0.30 \pm 0.04	3.53 \pm 0.58	12.7 \pm 2.16	19.5 \pm 1.18	5.71 \pm 0.54	0.68 \pm 0.12	3.91 \pm 0.37	14.6 \pm 2.02	6.97 \pm 0.99
<i>Salmea petroboides</i>	0.73	22.2 \pm 1.58	0.13 \pm 0.04	13.4 \pm 0.96	6.97 \pm 0.49	10.2 \pm 0.48	10.5 \pm 0.51	0.71 \pm 0.04	13.1 \pm 1.59	26.1 \pm 0.83	4.40 \pm 0.20
<i>Croton linearis</i>	0.83	3.00 \pm 0.33	0.32 \pm 0.02	15.0 \pm 0.95	20.0 \pm 1.26	7.68 \pm 0.49	10.8 \pm 0.71	1.00 \pm 0.18	8.16 \pm 0.79	12.1 \pm 2.57	2.50 \pm 0.31
Kruskal–Wallis H		33.2**	24.5*	41.5**	41.4**	42.6**	38.6**	32.6**	33.9**	23.7*	33.9**

^a SE is zero because only one plant was used to obtain this measurement.

overall leaf toughness across multiple species (Kitajima & Porter 2010, Westbrook *et al.* 2011), and our study supports this.

Toughness in leaves can affect iguanas by slowing foraging efficiency and by decreasing food digestibility. For any two given foods (other features of their loading in the mouth being similar), a tougher food will require more work per area to fracture during chewing (Wright & Vincent 1996). Investigations focusing on prey processing behaviour and jaw mechanics suggest that reptiles chew (Ross *et al.* 2010; but see Fritz *et al.* 2010) and iguanas often bite repeatedly once a piece of leaf has been sheared, or when leaves have been pulled away at the petiole. Digestion by microbes is facilitated by cuts or fractures in the cuticle (Bjorndal *et al.* 1990), and thus it is conceivable that digestion efficiency is increased via punctures from repeated bites that are deeper and more severe in a leaf that is relatively less tough. Additional research is needed to support this supposition (Bjorndal *et al.* 1990).

Our results offer a basis for future testing in herbivorous lizards and should be expanded to include influences of varying leaf morphological, chemical, and biomechanical traits resulting from seasonal effects and phenological stages of leaves. Iguanas consume seasonal fruits; therefore, expanding this study to consider fruits and flowers would further advance our understanding of food selection criteria in herbivorous lizards. Nonetheless, the results from this study are the first to demonstrate that physical leaf traits can influence food selection in a true herbivorous lizard and offer a testable hypothesis for further study.

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